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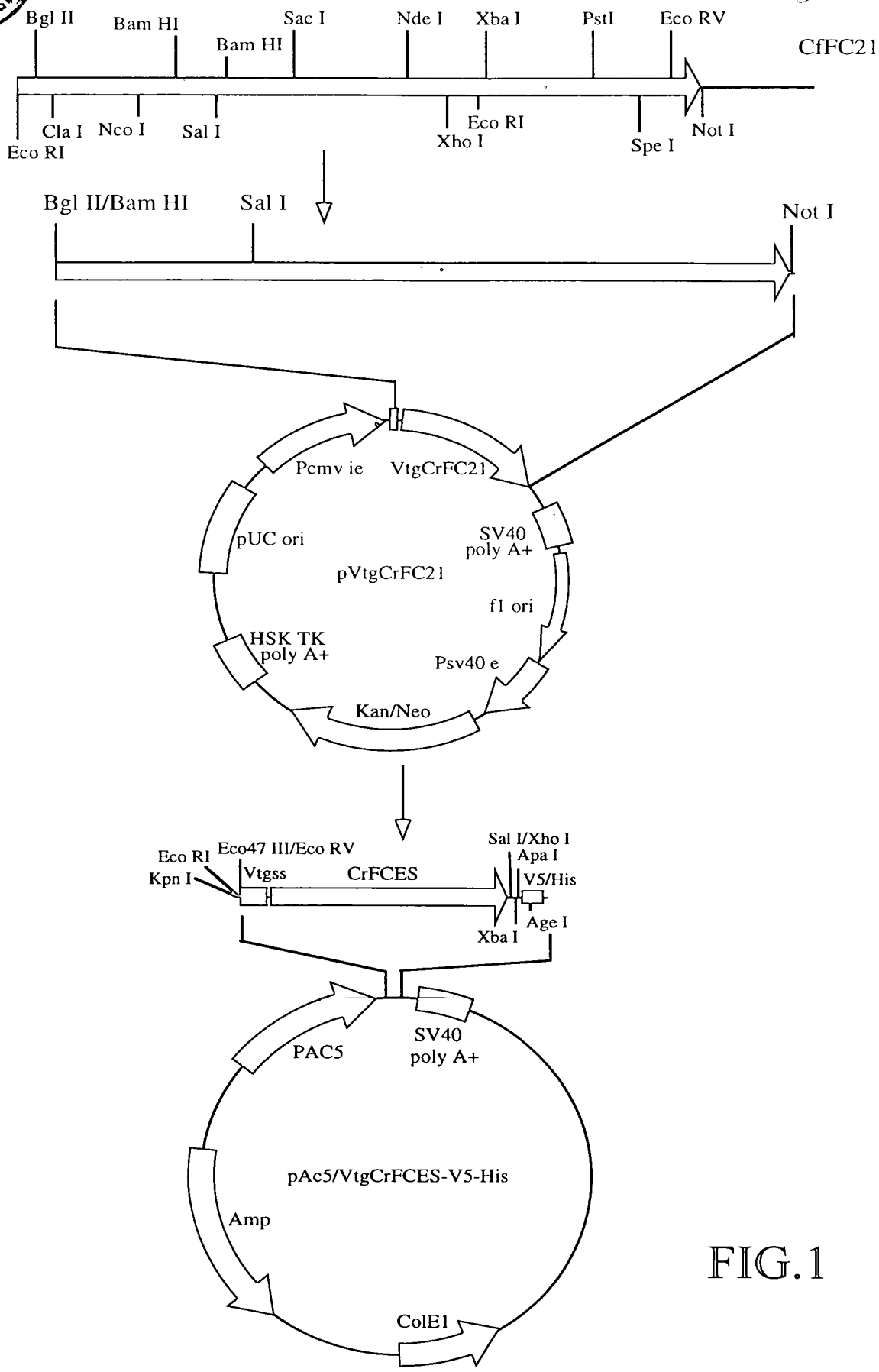
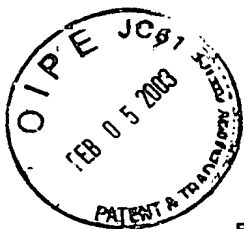


FIG.1



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5'
 GTG GAA TTC TGC AGA TGC TAC CGG ACT CAG ATC AAT TCA CAT CCA CCA GCC
 ATG AGG GTG CTT GTA CTA GCT CTT GCT GTG GCT CTC GCA GTG GGG GAC CAG
 M R V L V L A L A V A L A V G D Q
 OaVtgss → Cleavage site
 TCC AAC TTG GGG GAT CTA GGC TTG TGT GAT GAA ACG AGG TTC GAG TGT AAG
 S N L G D L G L C D E T R F E C K
 → Factor C
 TGT GGC GAT CCA GGC TAT GTG TTC AAC ATT CCA GTG AAA CAA TGT ACA TAC 3'
 C G D P G Y V F N I P V K Q C Y F

pAc5/VtgCrFCES-V5-His

FIG.2A

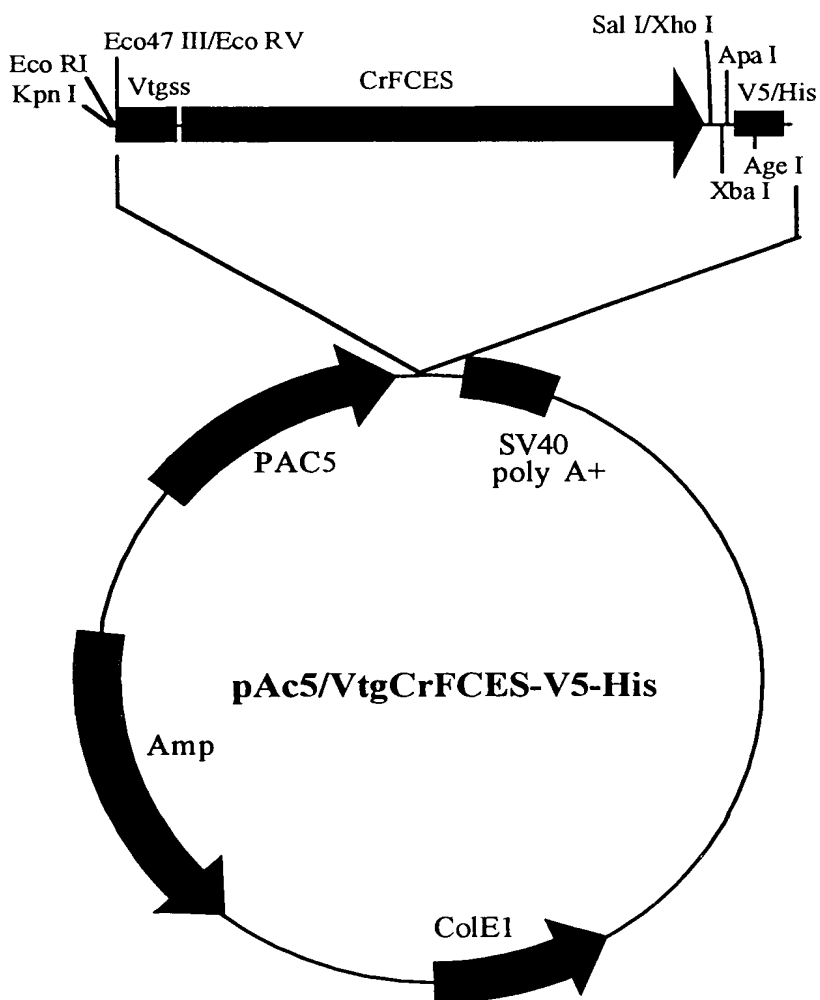


FIG.2B

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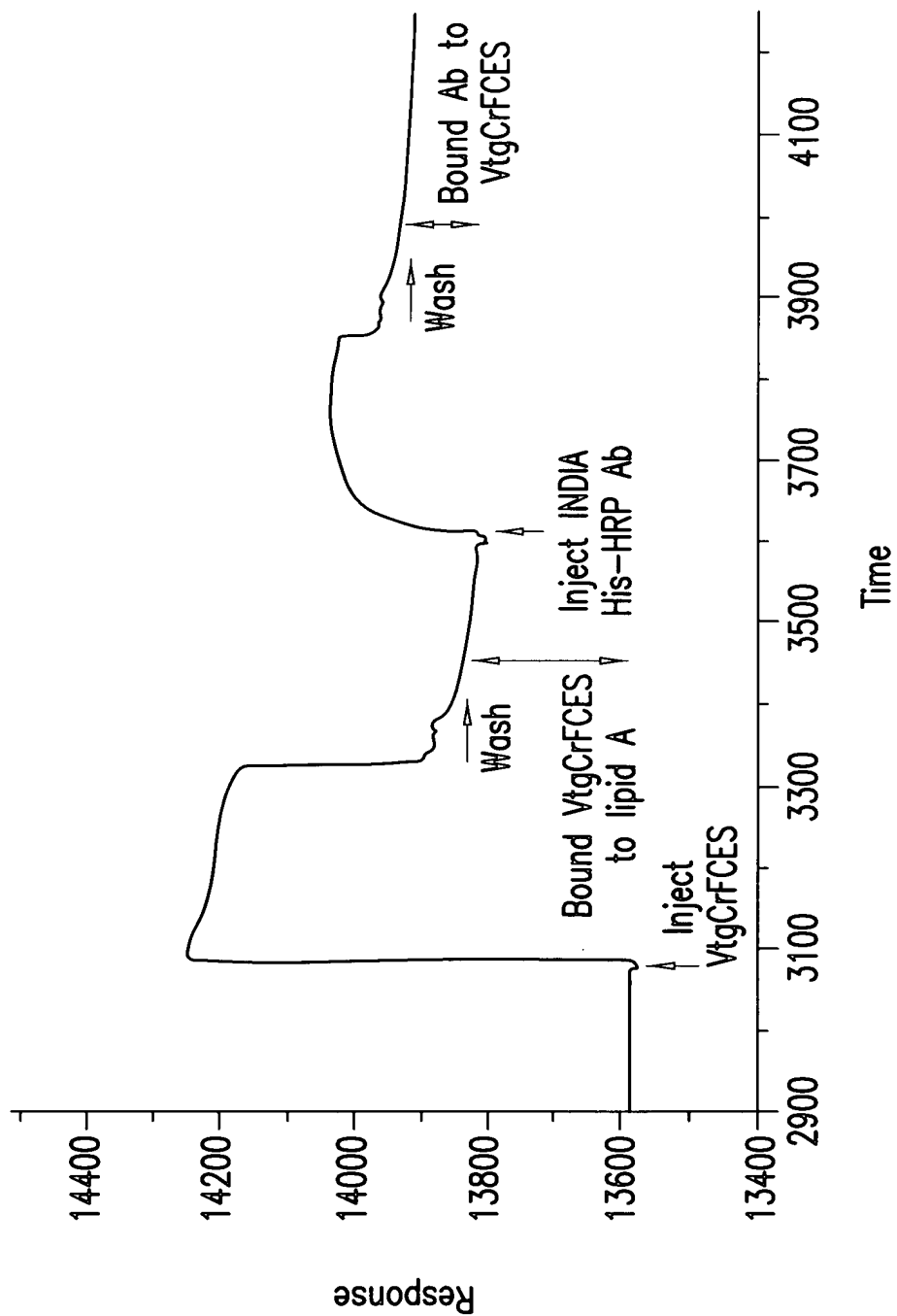


FIG.4



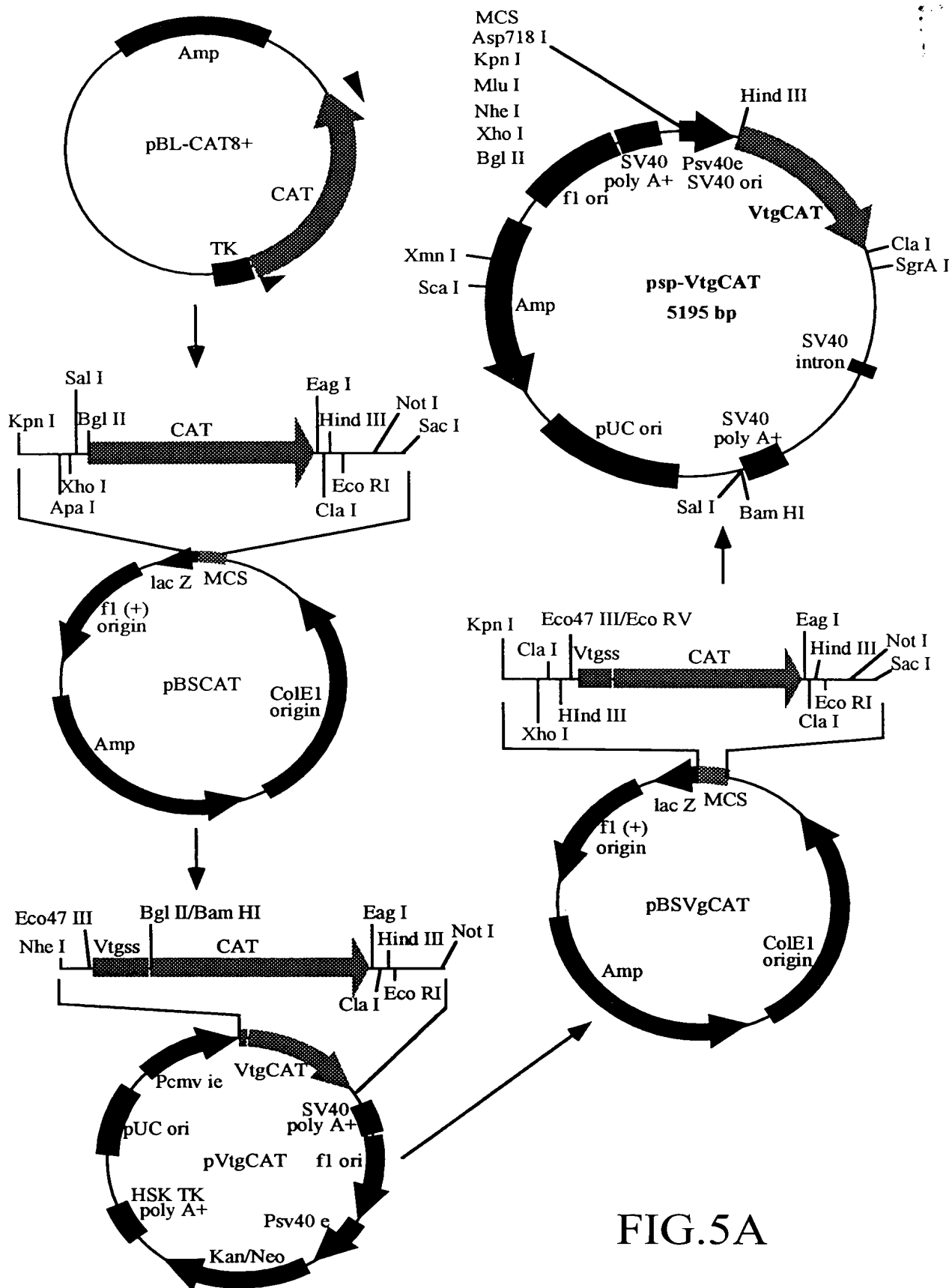


FIG.5A

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5'

Hind III Eco RV/Eco 47III
TGC AAA AAG CTT GAT GCT ACC GGA CTC AGA TCA ATT CAC ATC CAC CAG CC
ATG AGG GTG CTT GTA CTA GCT CTT GCT GTG GCT CTC GCA GTG GGG GAC CAG
M R V L V L A L A V A L A V G D Q
OaVtgss → Cleavage site

Bgl II/Bam HI
TCC AAC TTG GGG GAT CTG CTG GAG AAA AAA ATC ACT GGA TAT ACC ACC GTT
S N L G D L L Q K K V T G W T T V
CAT →

Eag I
... .. GGC GGG GCG TAA TTT TTT TAA GGC ACG GCC GAT GCG ACG
G G A ***

Cla I
GTA TCG ATA TTG TTA CAA CAC CCC AAC 3'

psp-VtgCAT

FIG.5D

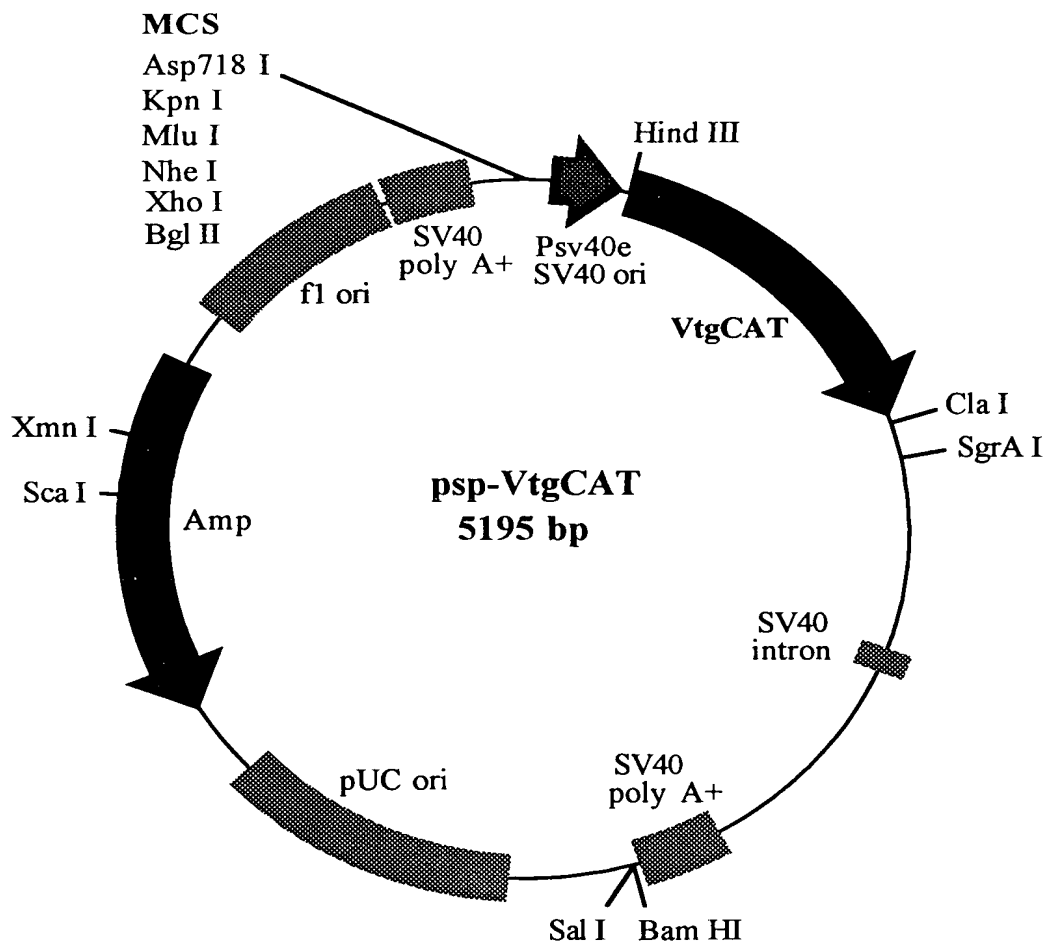


FIG.5E

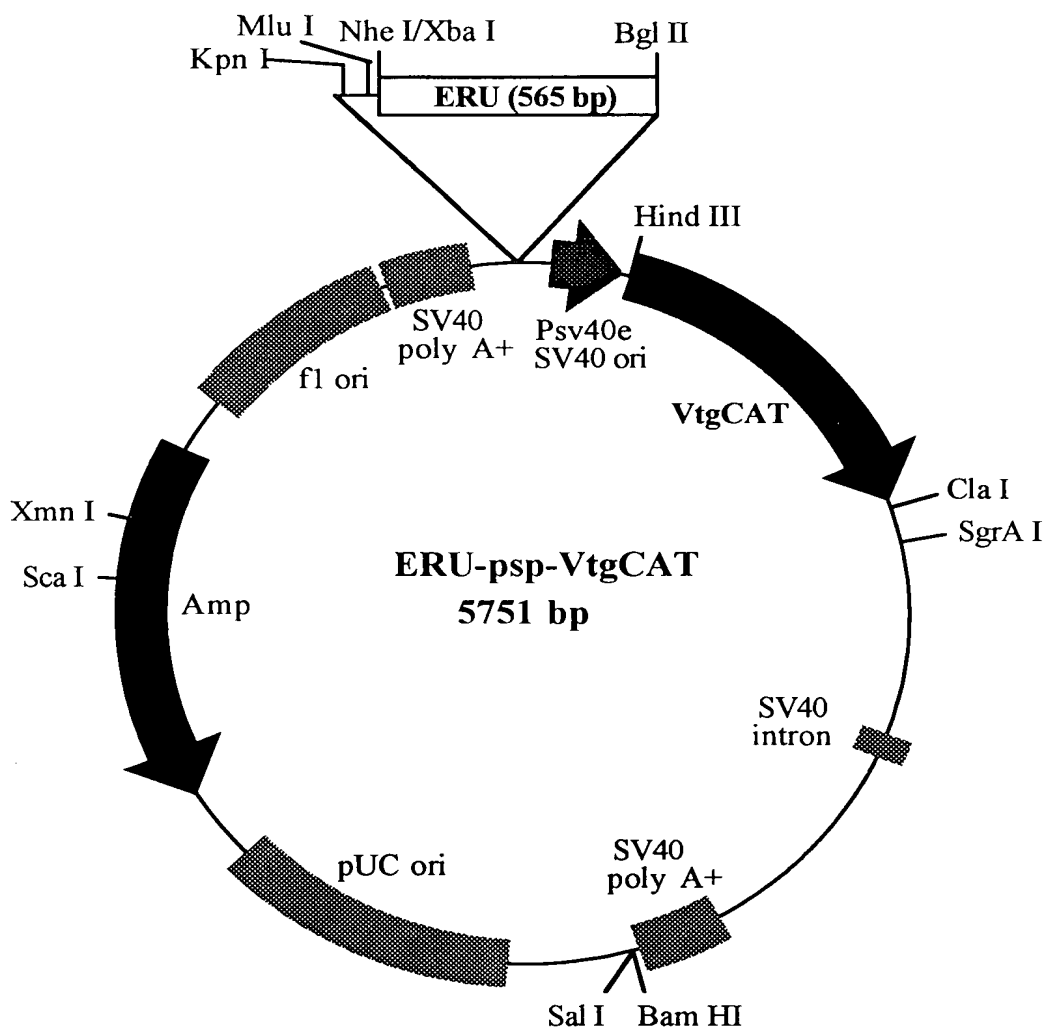


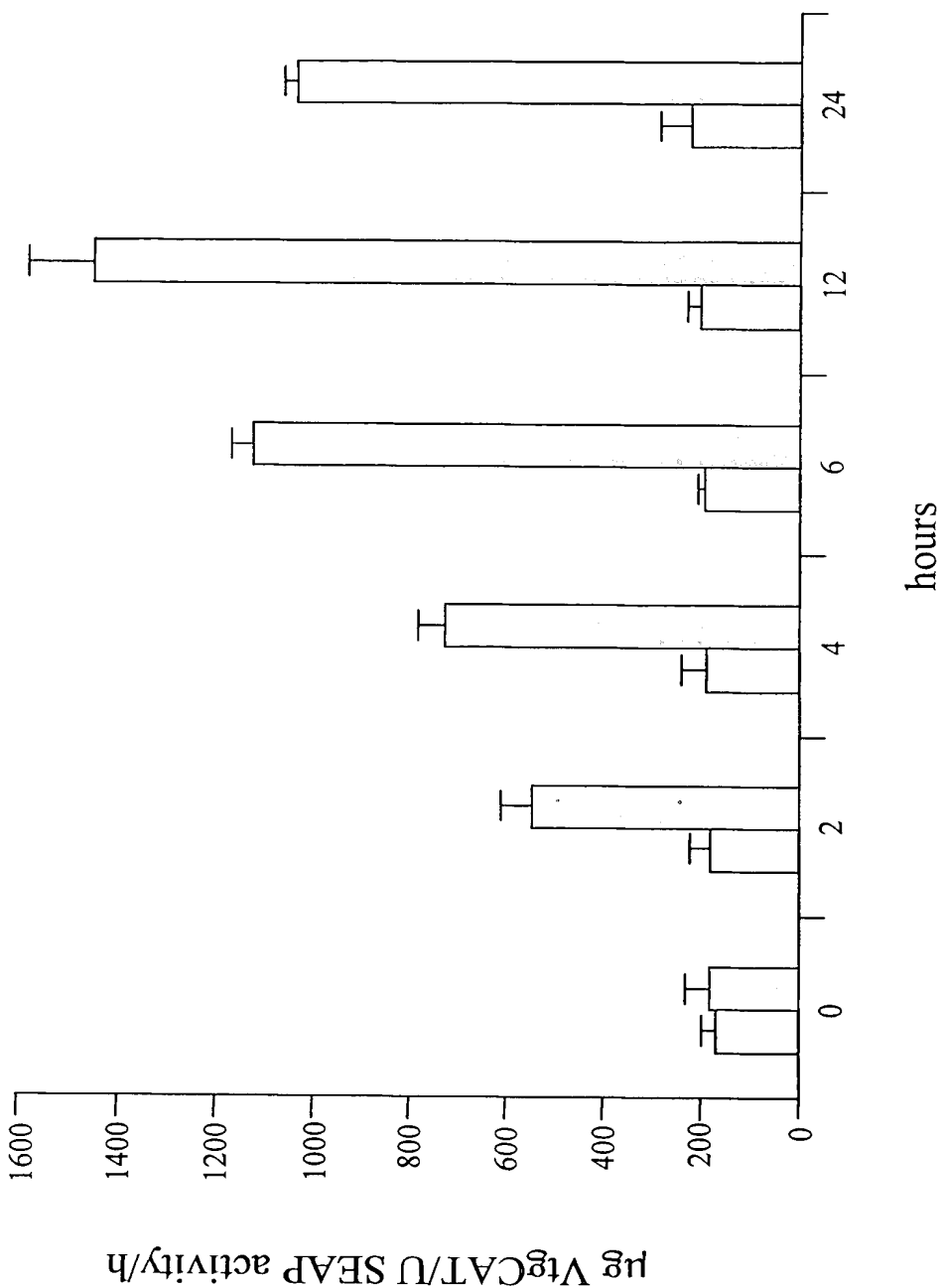
FIG.5F

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FIG.6



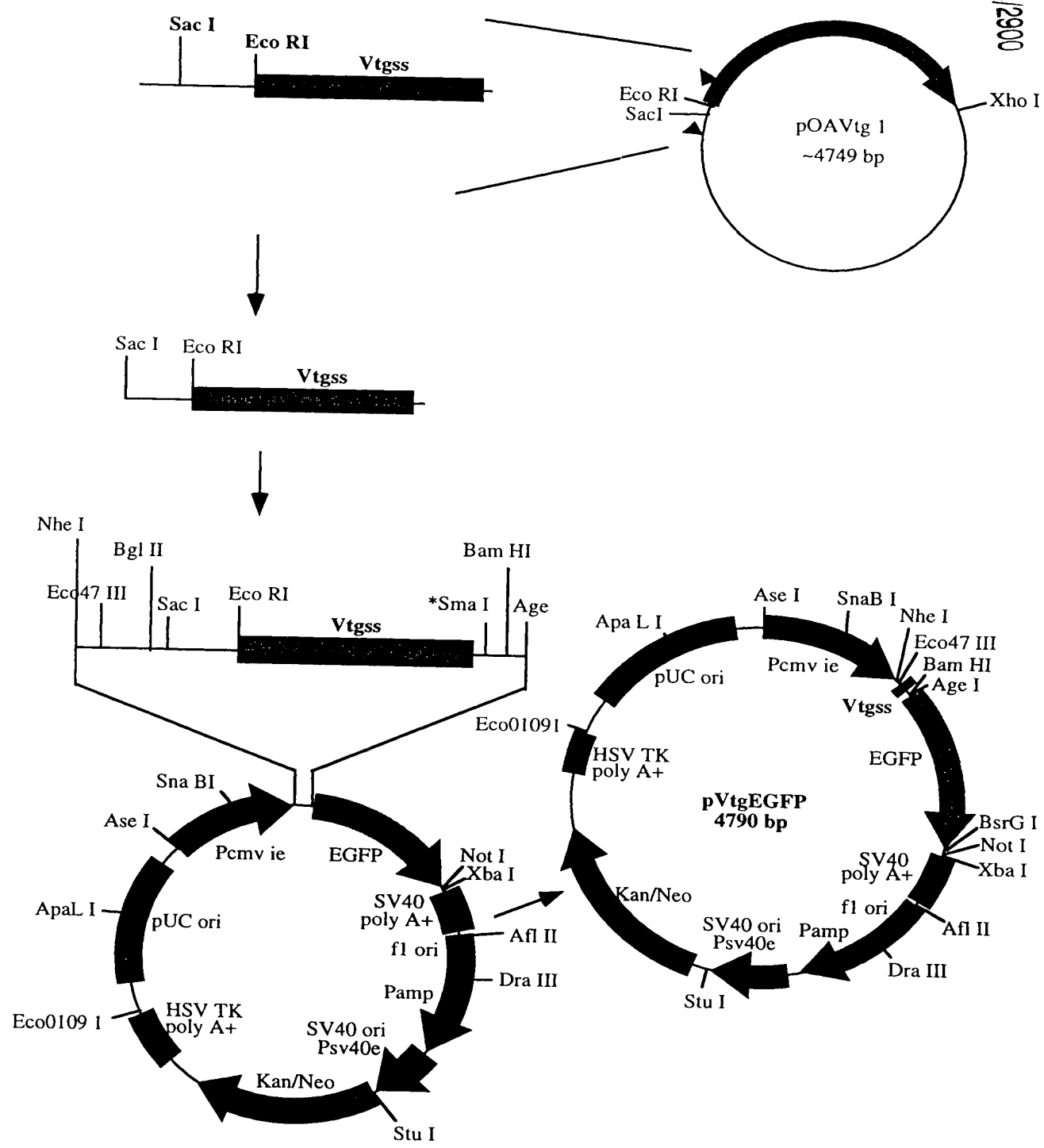


FIG.8A

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5'

Nhe I

+1

GCT AGC GCT ACC GGA CTC AGA TCA ATT CAC ATC CAC CAG CC

Eco 47 III

ATG AGG GTG CTT GTA CTA GCT CTT GCT GTG GCT CTC GCA GTG GGG GAC CAG
 M R V L V L A L A V A L A V G D Q

OaVtgss →Bam HI Age I

TCC AAC TTG GGG GAT CCA CCG GTC GCC ACC ATG GTG AGC AAG GGC GTG GTG
 S N L G D P P V A T M V S K G V V

→ EGFP

CAG AAC TCC GGG 3'

Q N S G

pVtgEGFP

FIG.8B

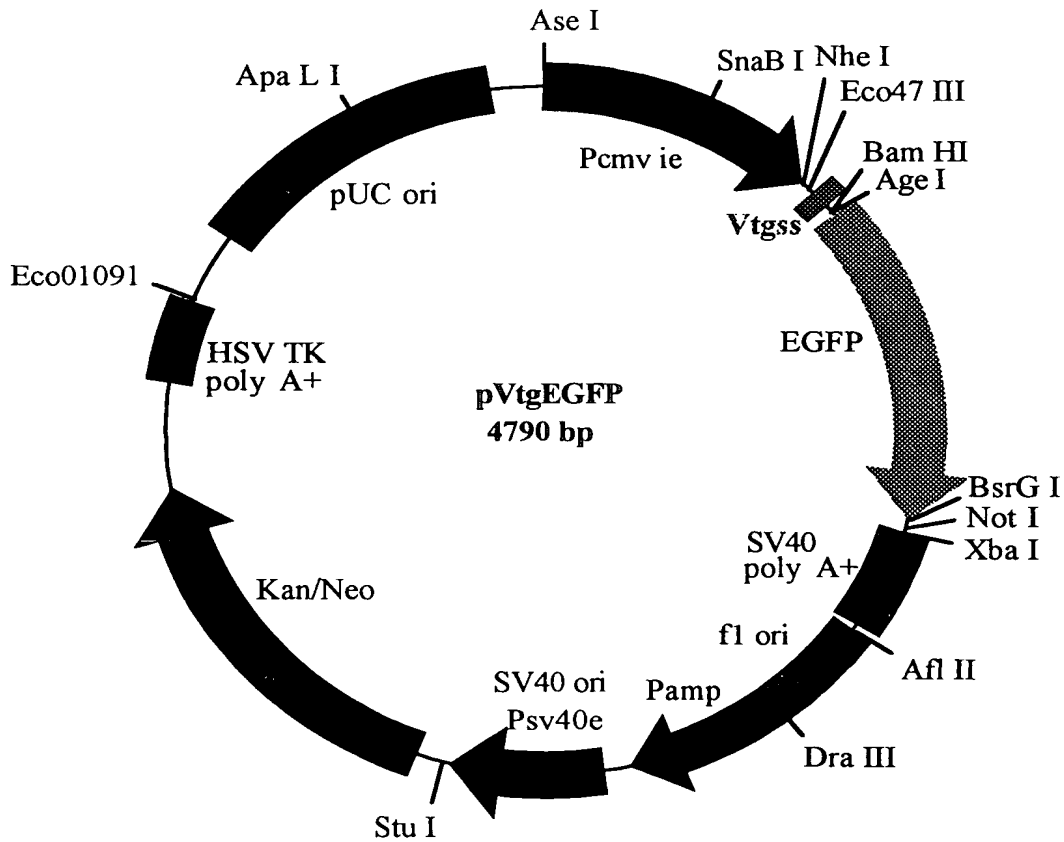


FIG.8C

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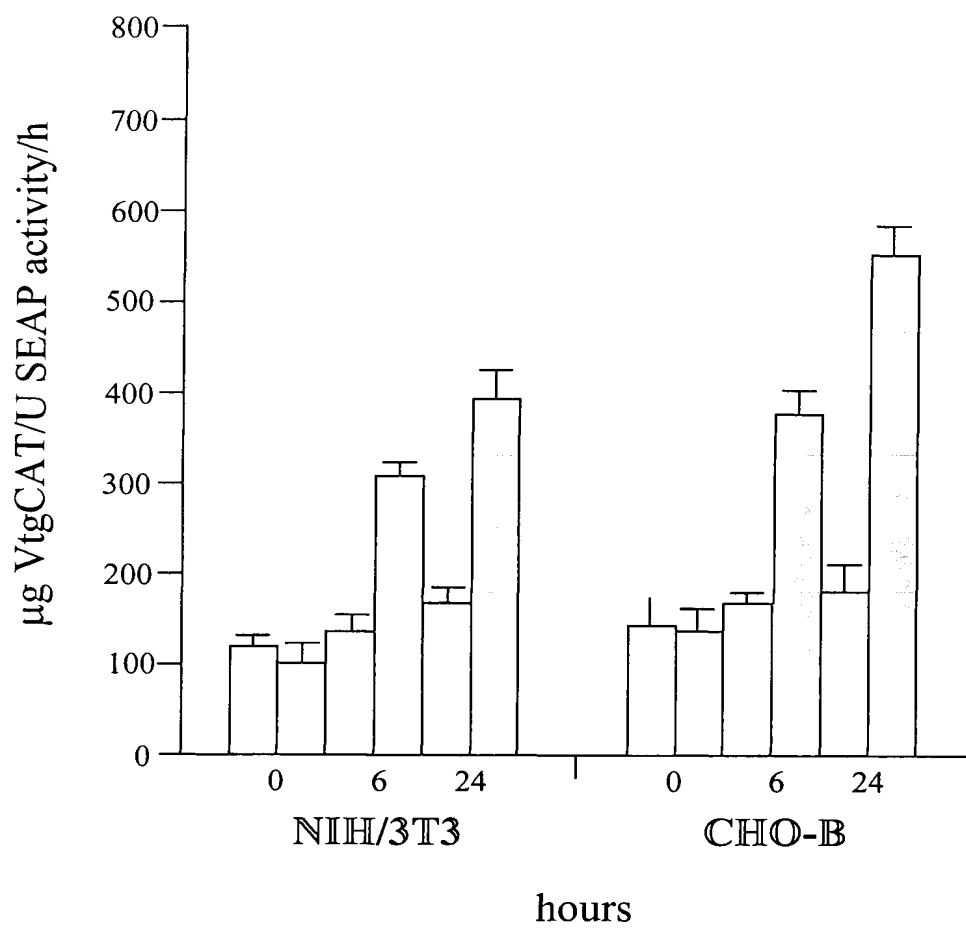


FIG.9

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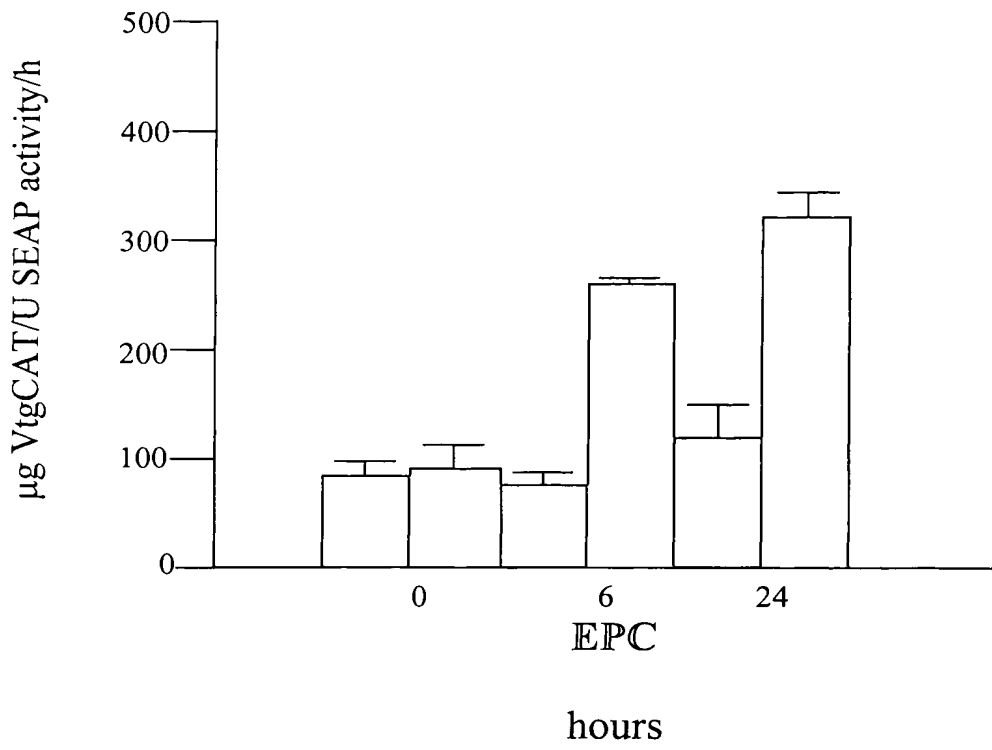


FIG.10



Expression profile of VtgCAT in
two different yeast transformants over 72 hours

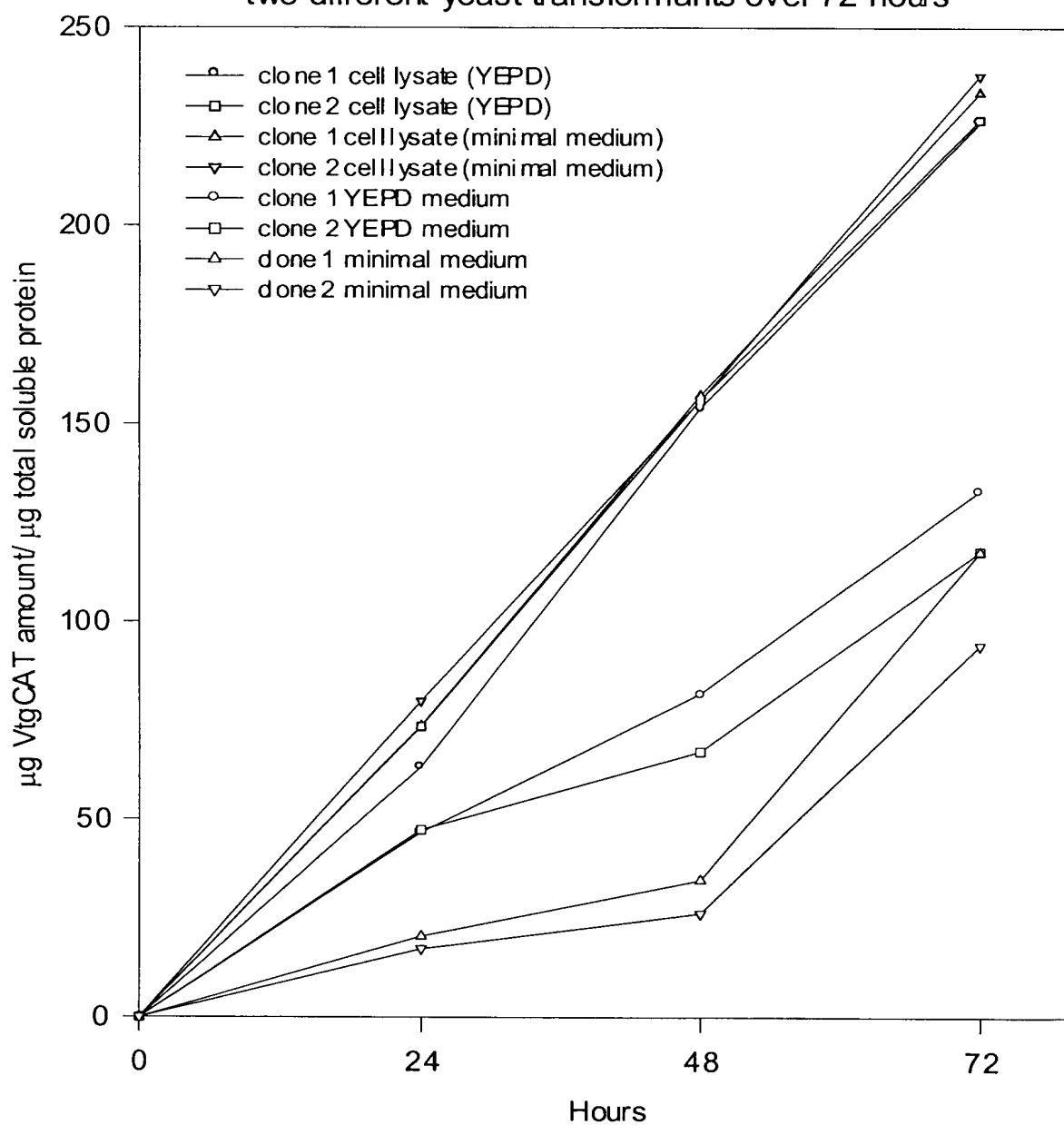


FIG.12

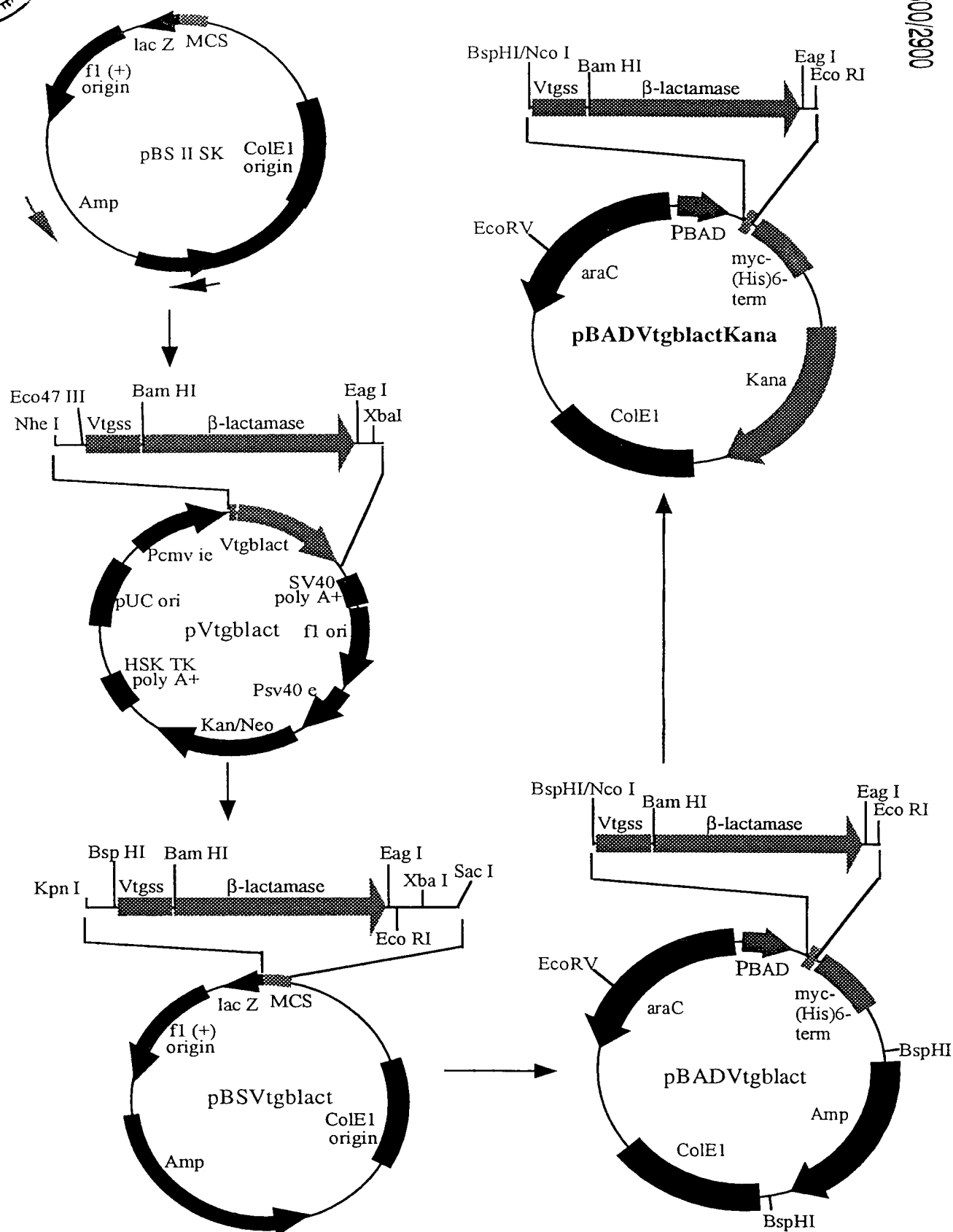
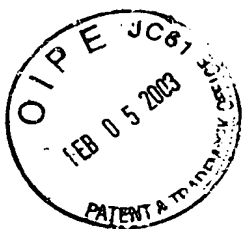
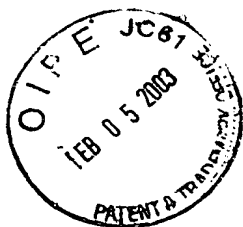


FIG.13



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5' BspHI/NcoI
 CTC TAC TGT TTC TCC ATA CCC GTT TTT TTG GGC TAA CAG GAG GAA TTA ACC
 ATG AGG GTG CTT GTA CTA GCT CTT GCT GTG GCT CTC GCA GTG GGG GAC CAG
 M R V L V L A L A V A L A V G D Q
 OaVtgss → Cleavage site
Bam HI
 TCC AAC TTG GGG GAT CCA GAA ACG CTG GTG AAA GTA AAA GAT GCT GAA GAT
 S N L G D P E T L V K V K D A E D
β-lactamase
 CAG TTG GGT GCA CGA GTG GGT TAC ATC GAA CTG GAT CTC AAC AGC GGT AAG 3'
 Q L G A R V G Y I E L D L N S G K

pBADVtgblactKana

FIG.14A

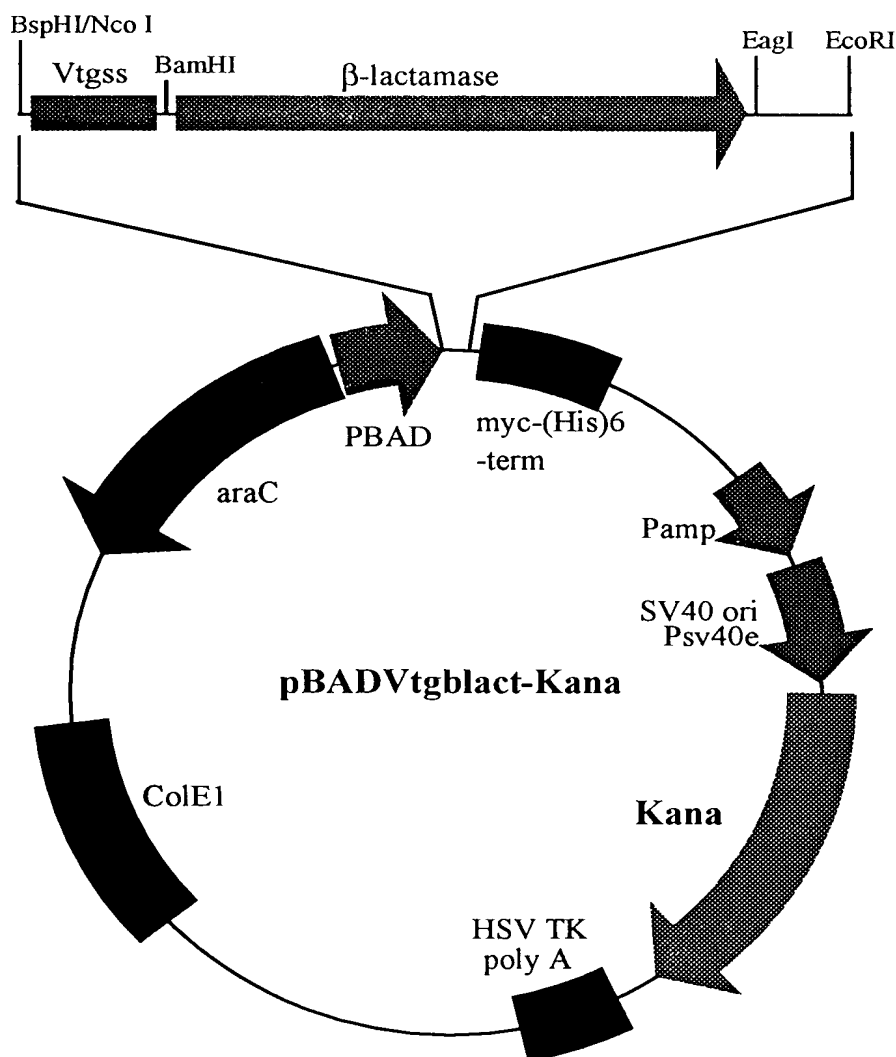


FIG.14B



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Growth profile of bacteria after induction

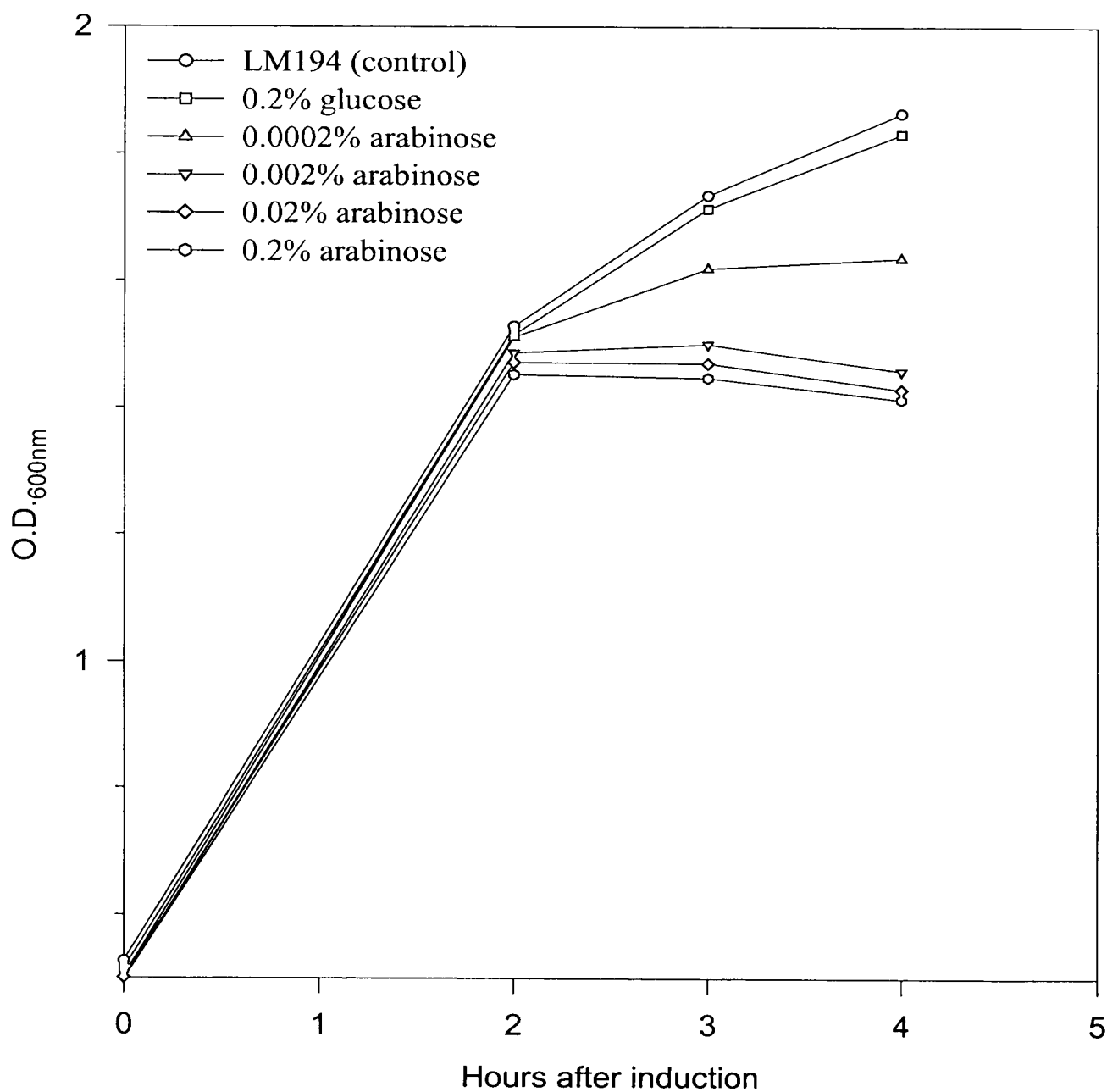
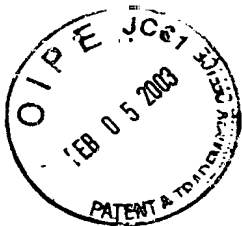


FIG.16



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β -lactamase in periplasmic space over time

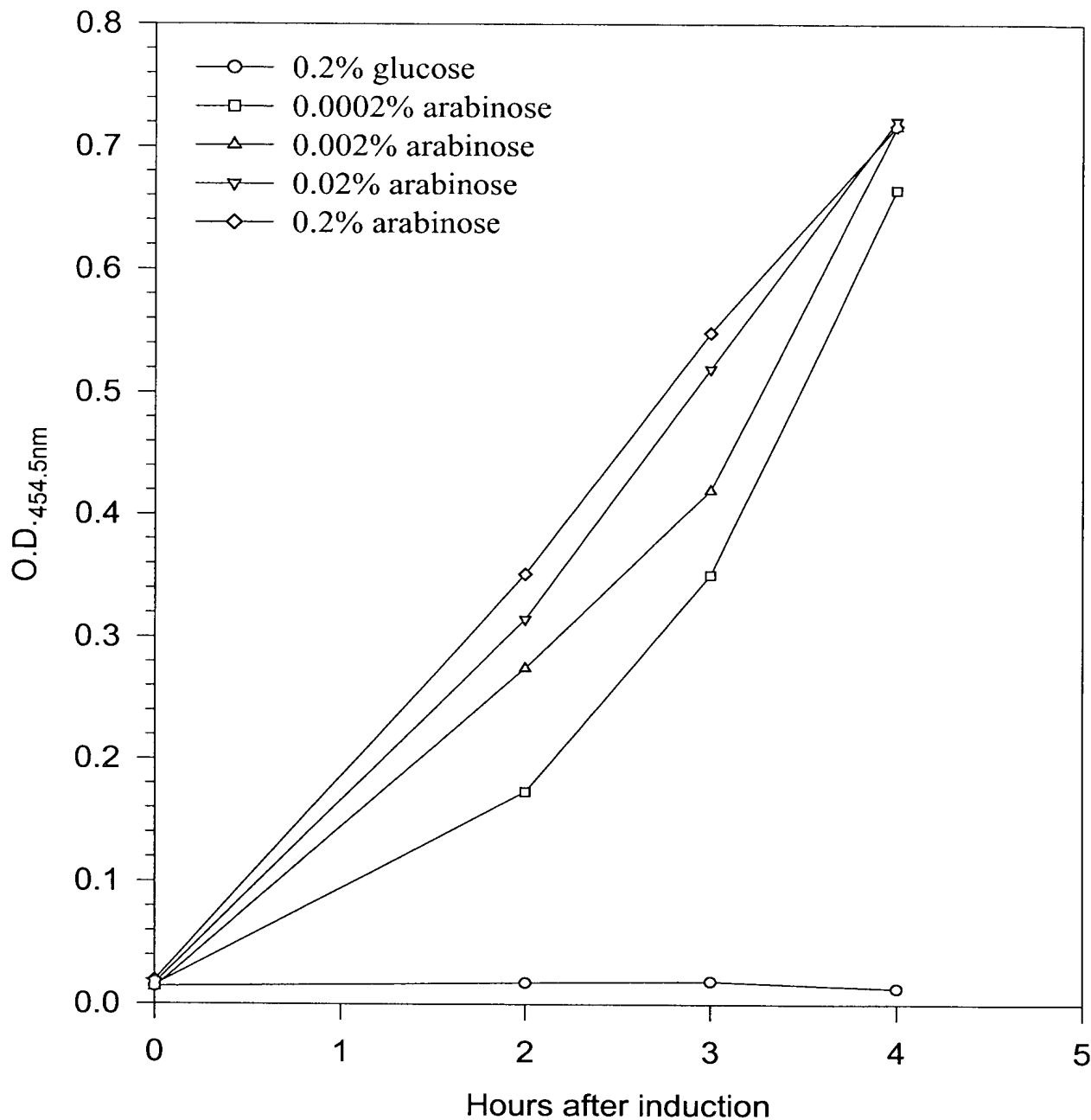


FIG.17



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β -lactamase in culture medium over time

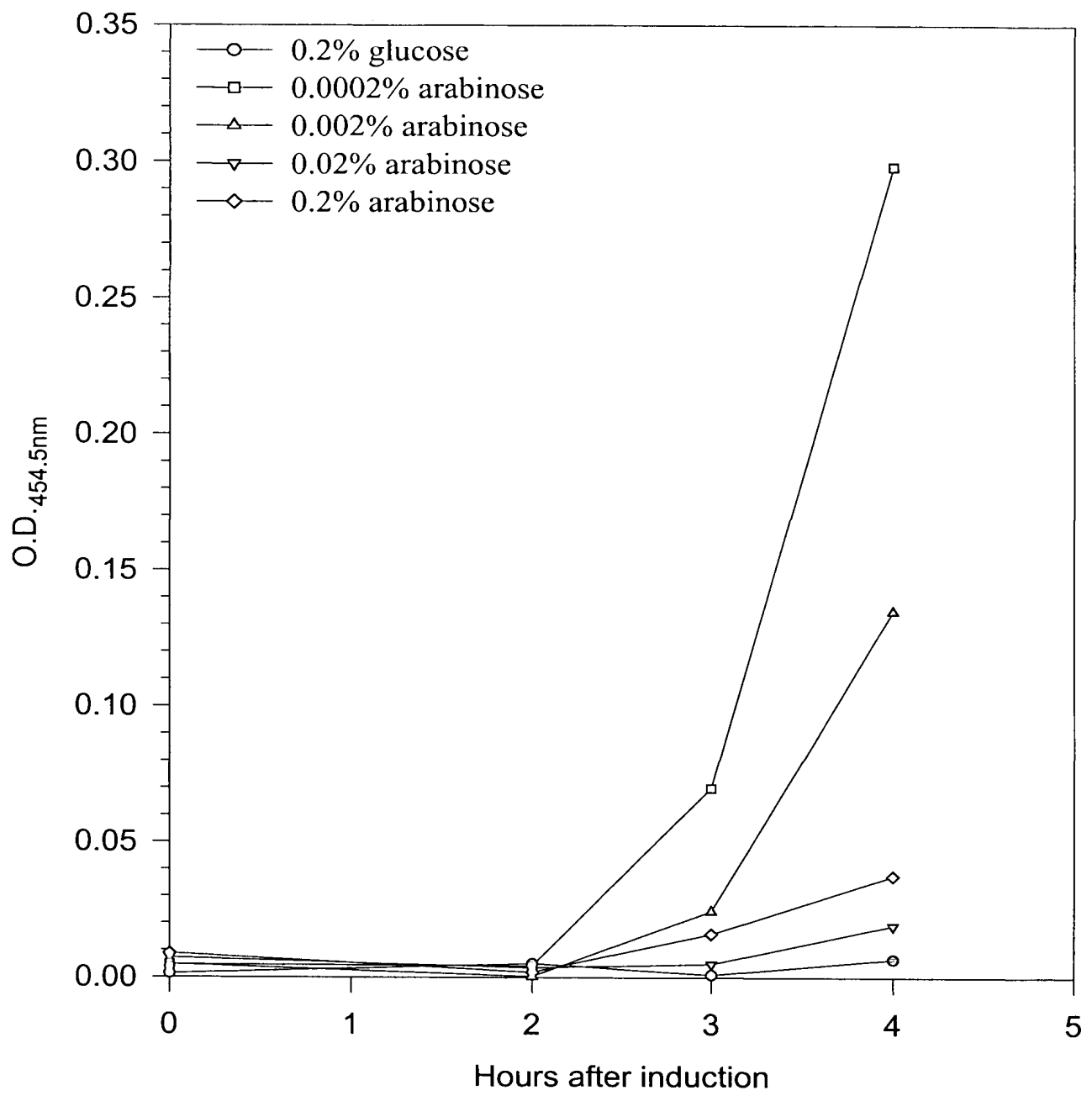


FIG.18

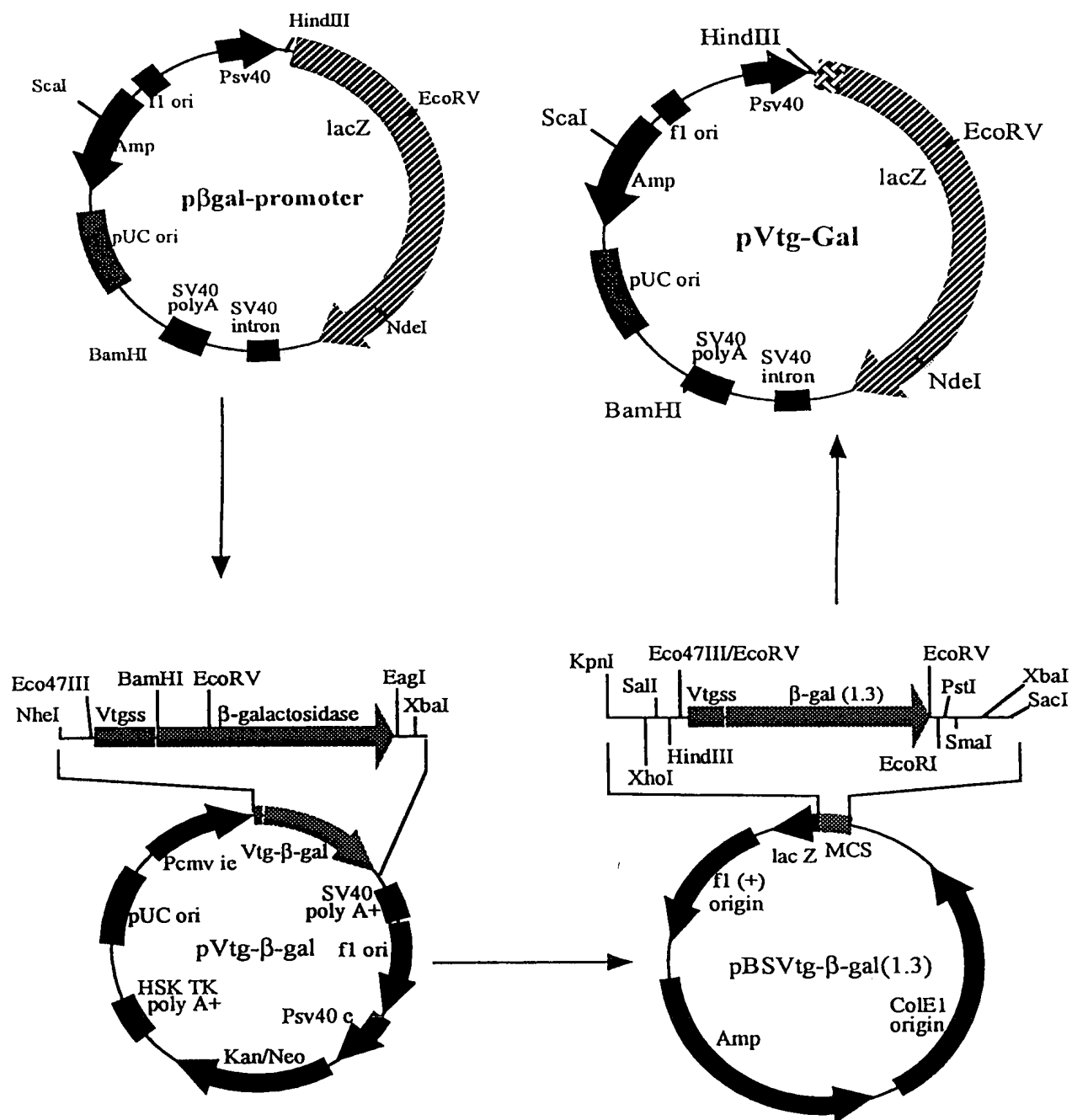


FIG.19

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Rate of Secretion

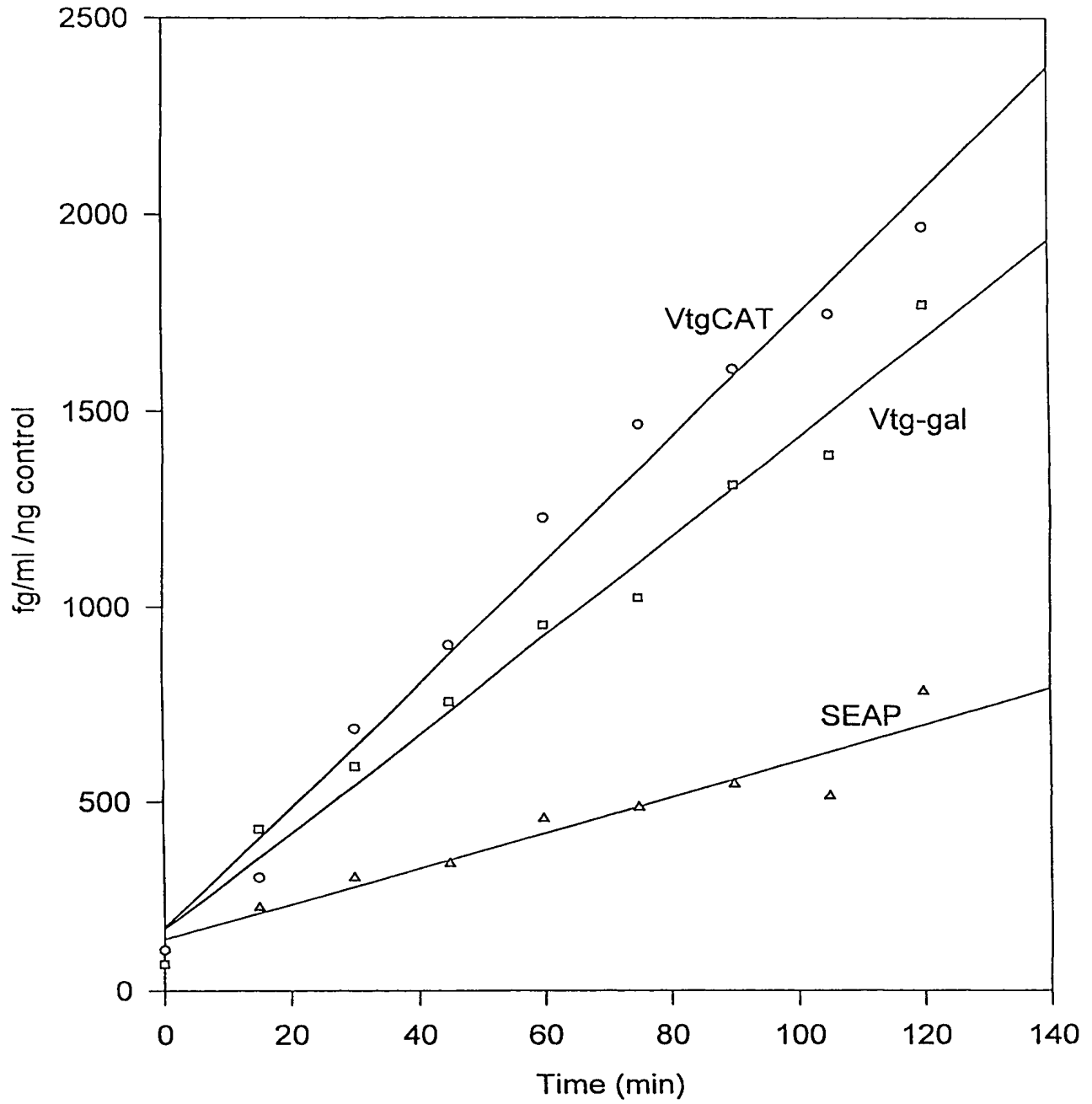


FIG.21



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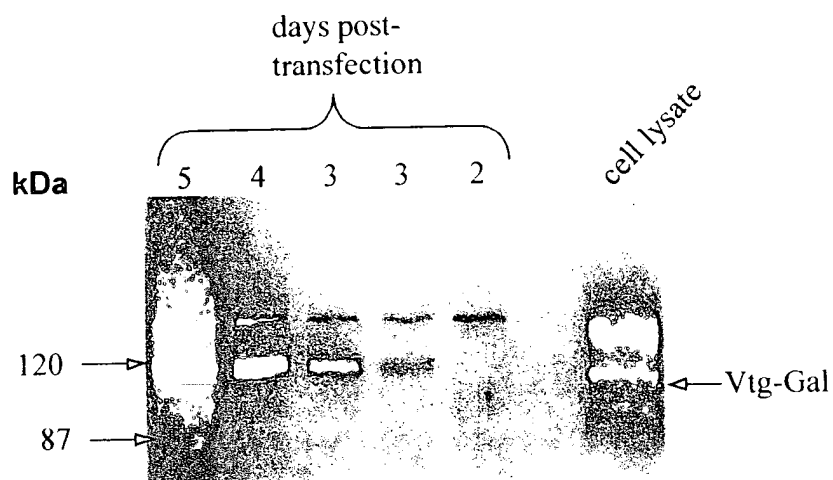
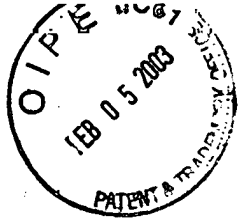


FIG.20



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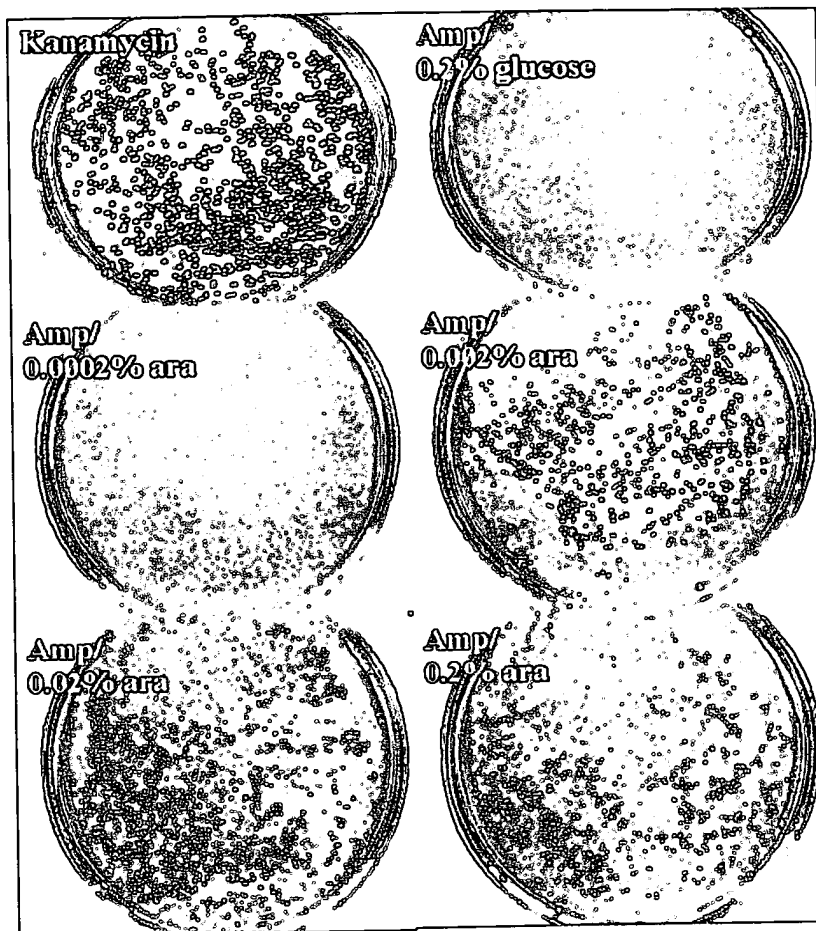


FIG.15



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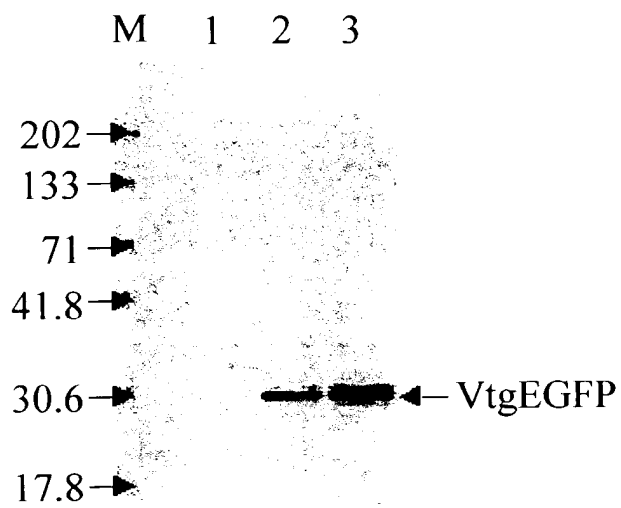
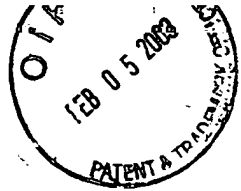


FIG.8D



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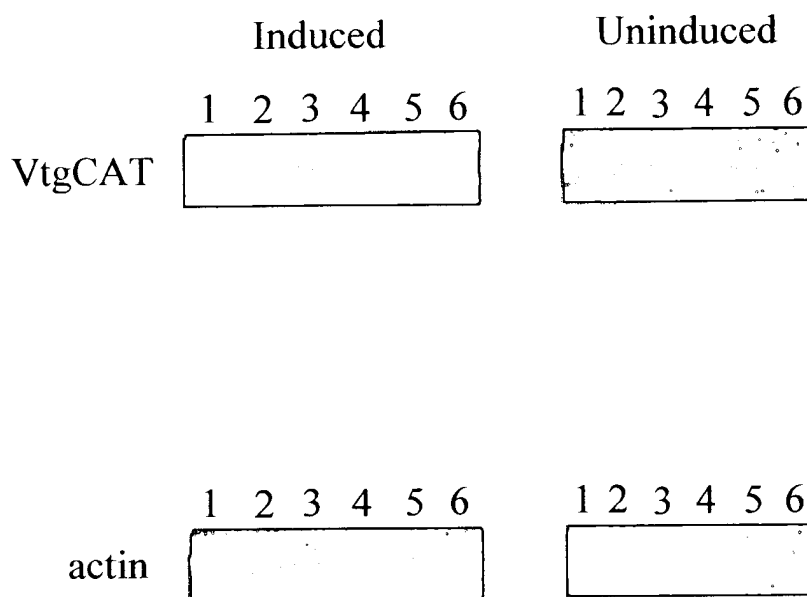


FIG.7

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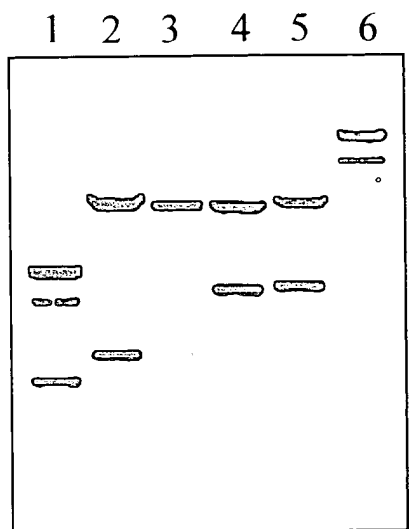
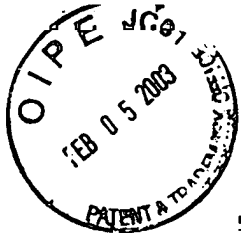


FIG.5G



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5' Hind III Eco RV/Eco 47III
ATC GAT AAG CTT GAT GCT ACC GGA CTC AGA TCA ATT CAC ATC CAC CAG CC

ATG AGG GTG CTT GTA CTA GCT CTT GCT GTG GCT CTC GCA GTG GGG GAC CAG
M R V L V L A L A V A L A V G D Q
OaVtgss → Cleavage site

Bgl II/Bam HI
TCC AAC TTG GGG GAT CTG CTG GAG AAA AAA ATC ACT GGA TAT ACC ACC GTT
S N L G D L L Q K K V T G W T T V
CAT →

Eag I
... GGC GGG GCG TAA TTT TTT TAA GGC ACG GCC GAT GCG ACG
G G A ***

Cla I
GTA TCG ATA ACT TGA TAT CG 3'
Hind III

pBSVtgCAT

FIG.5B

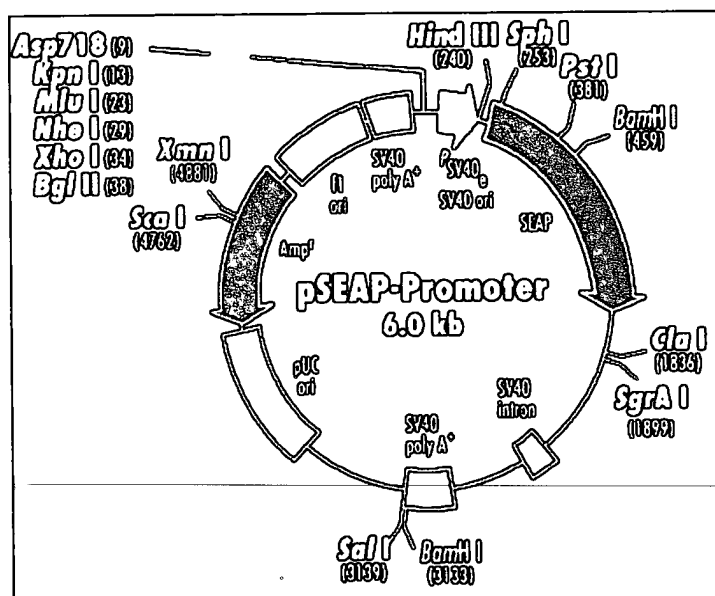


FIG.5C



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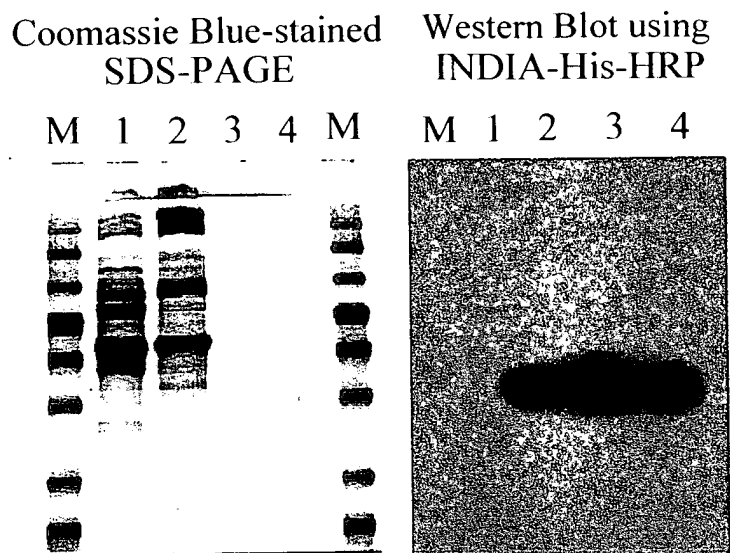


FIG.3A

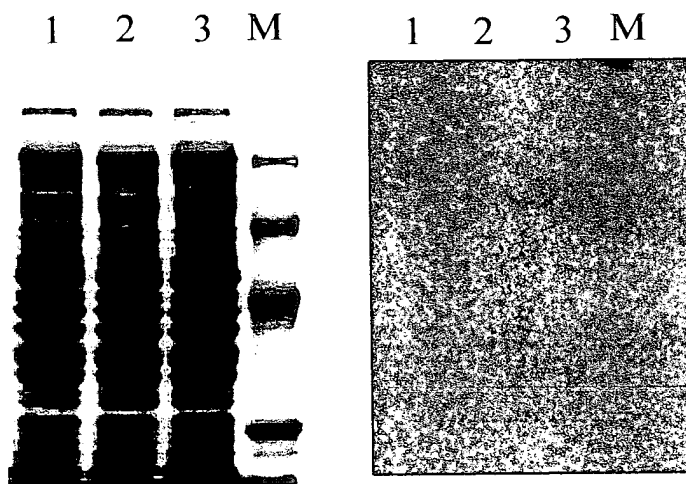


FIG.3B